Effect of Imidacloprid Soil Treatments on Occurrence of Formosan Subterranean Termites (Isoptera: Rhinotermitidae) in Independent Monitors

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ABSTRACT Periodic sampling of 30 independent monitors, initially active with the Formosan subterranean termite, *Coptotermes formosanus* Shiraki, was conducted to evaluate the effects of soil treated with imidacloprid on nearby termite activity. Monitors were located adjacent (1–3 m) to the buildings. Soil around and under the buildings was treated with 0.05% imidacloprid. None of the termites collected showed latent mortality attributed to imidacloprid intoxication. Imidacloprid soil treatments did not measurably reduce *C. formosanus* populations adjacent to the treatments. Imidacloprid does not seem to fit the liquid-bait model.

KEY WORDS *Coptotermes formosanus*, termite, imidacloprid, dwellings

TOTAL ECONOMIC LOSS DUE to termites in the United States was estimated at $11 billion/yr (Su 2002). The Formosan subterranean termite, *Coptotermes formosanus* Shiraki, is native to Asia (Bouillon 1970), but it was introduced into the southern United States where it has become a devastating pest (Su and Tamashiro 1987). In addition to structural infestations, *C. formosanus* infestations of living trees are common in the New Orleans, LA, area (Osbrink et al. 1999, 2001). Control of termite populations is critical because of the danger of their destroying urban structures.

It has been suggested that soil treatments with a slow-acting, nonrepellent insecticide would result in elimination of termite populations in areas adjacent to the treatments (Thorne and Breisch 2001, Potter and Hillery 2002). Such a liquid-bait would be very desirable because it would provide population suppression by eliminating colonies one by one but have advantages over a bait with the reduced labor of a soil termiticide. One proposed slow-acting, nonrepellent termiticide is the new generation neonicotinoid imidacloprid (Matsuda et al. 2001). Imidacloprid, a nicotine analog, is an insect-specific agonist of nicotinic acetylcholine receptors that has been reported to be nonrepellent to termites (Matsuda et al. 2001, Thorne and Breisch 2001).

The objective of this research was to determine whether treatment of soil around structures with this new generation, slow-acting, nonrepellent insecticide would control *C. formosanus* populations in areas adjacent to those treatments, thus fulfilling the requirements of the liquid-bait model.

This article reports the results of research only. Mention of a proprietary product does not constitute an endorsement or recommendation by the USDA for its use.

Materials and Methods

Independent Monitors. In 1999, the ≈40-ha University of New Orleans Lakefront campus in Orleans Parish, LA, was surveyed for subterranean termites with placement of pine stakes (2 by 4 by 20 cm). This resulted in the establishment of 57 bucket trap termite monitors (Su and Scheffrahn 1986) active with *C. formosanus* located at a distance of 1–3 m around the perimeter of eight buildings (Fig. 1). Termites were identified from Scheffrahn and Su (1994).

In May 2001, pest management professionals treated the soil under and around four of the buildings (Fig. 1) with 0.05% imidacloprid (Premise 75, Bayer, Kansas City, MO). Treated buildings were Administration (Ad.); Cove, University Center (U.C.); and Chemical Annex (C.A.). The other four buildings were left untreated and used as controls, including Biology (Biol.), Dormitory (Dorm.), Engineering (Eng.), and Liberal Arts (L.A.). The number of monitors per building and the sampling regime are reported in Tables 2 and 3.

Beginning November 2000, *C. formosanus* were collected from monitors approximately monthly and maintained in the laboratory on stacked, moistened spruce (*Picea* sp.) slats (10 by four by 0.5 cm) in plastic containers (13 by 13 by 4 cm) at ≈100% RH and ≈27°C. Healthy termites fed, produced carton material, sought harborage, and survived for months. Termites intoxicated with imidacloprid would not feed, produce carton material, seek harborage, or survive beyond 14 d (Osbrink and Lax 2003).

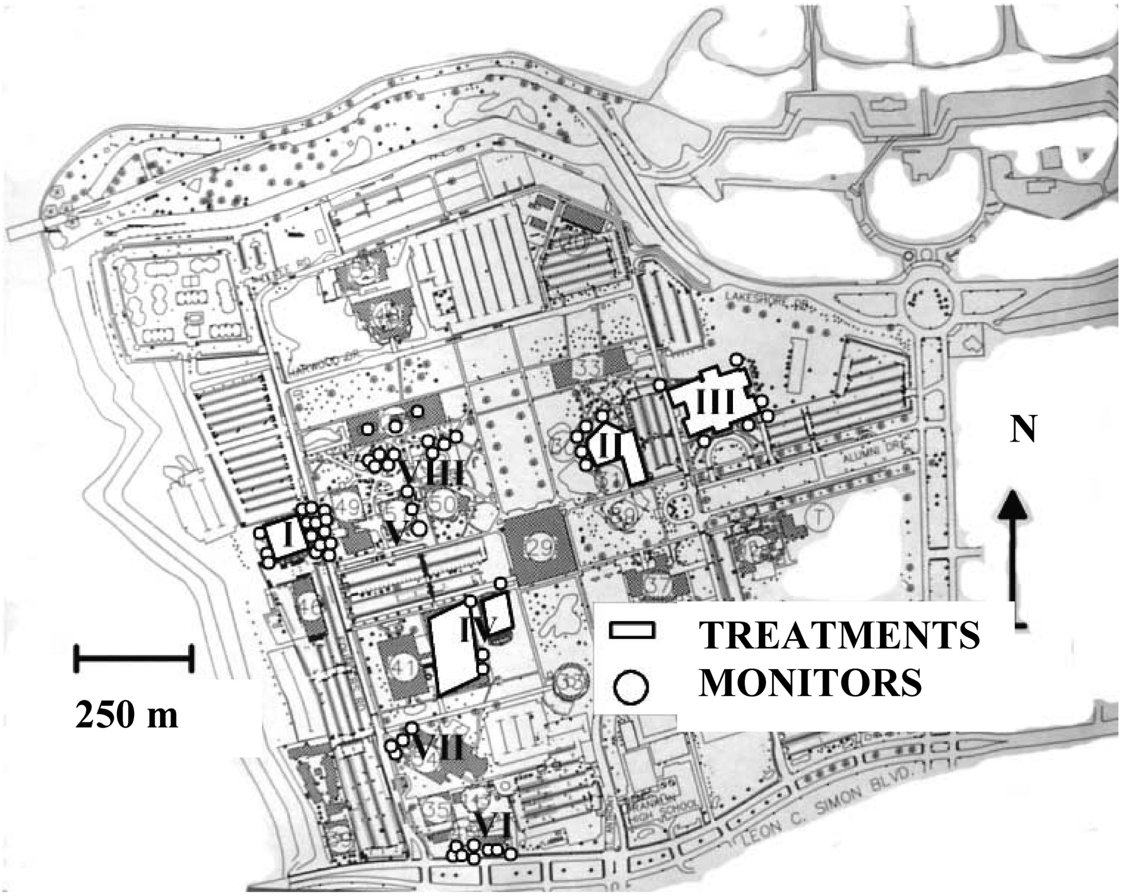

Fig. 1. Map of University of New Orleans indicating locations of *C. formosanus* traps (open circles) and treated buildings (open polygons).

Number of workers collected from each monitor was estimated by subtracting the total weight of soldiers and brachypterous nymphs from the total weight of collected termites (individual weights calculated by weighing four groups of 10 workers, soldiers, and brachypterous nymphs). Mean number of workers per building per collection date (from July 2000 to July 2002) was calculated by dividing number of workers collected by number of monitors per building and analyzed using analysis of variance (ANOVA) (PROC GLM, SAS Institute 1990). Means were separated using a least significant difference (LSD) test ($P < 0.05$; PROC GLM, SAS Institute 1990). Proportion of monitors with termites per building per collection date (from June 2001 to December 2001) was calculated by dividing the number of monitors with termites by the total number of monitors around a particular building, transformed by arcsine square root, and analyzed using ANOVA (PROC GLM, SAS Institute 1990). Means were separated using a LSD test ($P < 0.05$; PROC GLM, SAS Institute 1990) with the actual proportion reported in the tables. Posttreatment mean proportion of monitors with termites, by building, all dates combined, was transformed by arcsine square root and analyzed by ANOVA with means separated using a LSD test ($P < 0.05$; PROC GLM, SAS Institute 1990) with actual proportion reported in table.

Soil Residues. Soil was collected ≈1 yr posttreatment (May 2002) with a LaMotte (Chestertown, MD) soil sampler (2.54 cm i.d. by 25.4 cm in height) to a depth of ≈15 cm at a distance ≈15 cm from the foundation of each side of the four treated structures to obtain ≈1 liter of soil for each structure. Soil from the different sides of a specific structure was mixed uniformly. Soil was held in mason jars in the dark at room temperature (26.7 ± 1°C). An additional treatment of soil from the Cove, spiked with 50 ppm (wt: wt) imidacloprid, was included. Spiked soil was prepared by dissolving imidacloprid in 150 ml of acetone, which was used to cover 130 g of substrate in a mason jar. The acetone was allowed to evaporate from the substrate with periodic stirring over a period of 7 d. Soil samples were evaluated for pesticide residues by Department of Agricultural Chemistry, Agricultural Experiment Station, Louisiana State University. Additional residue analysis of soil samples collected at this same time was conducted on October 2002. Soil residues were evaluated for the presence of the following chemicals: imidacloprid, permethrin, cypermethrin, fipronil, chlorpyrifos, and chlordane.

Standard operating procedures for imidacloprid extraction was accomplished with an acetonitrile shake and analyzed for imidacloprid on high-performance liquid chromatography. For imidacloprid, a soil sample was prepared by drying it on a sheet of aluminum foil under a hood and then grinding. To a 25-g soil sample was added 100 ml of acetonitrile, shaken on a mechanical shaker for 2 h or overnight. The sample was filtered into a flask, and the flask was rinsed with acetonitrile as needed. Volume was reduced on a water bath to ≈2 to 3 ml. The detection instrument was a chromatograph consisting of a Waters 600E four-solvent pumping system with a Waters 717 auto sampler and a Waters 996 photodiode array detector (Waters, Milford, MA). The mobile phase was 60:40 acetonitrile/water isocratic. Flow was 1.5 ml/min. The column was an ODS-2 (C18) with a UV 270-nm wavelength detector. Column temperature was ambient with an injection amount of 20 μl. The analytic lab included one spiked sample of 25 g of soil extracted in the same manner after 1-ml spiking level at 10 times the working standard level.

Samples were extracted for permethrin, cypermethrin, fipronil, chlorpyrifos, and chlordane on a Dionex ASE by using ethyl acetate. A gas chromatograph with an electron capture detector (GC-ECD) was used for chlordane, fipronil, chlorpyrifos, permethrin, and cypermethrin. GC-NPD (nitrogen phosphate detector) was used to clarify chlorpyrifos analysis. GC-MSD (mass spectrophotometer) was used for confirmation of chlordane and permethrin and to confirm the absence of fipronil and chlorpyrifos. The gas chromatograph was a Hewlett–Packard model 6890, auto sampler #7683. Chem Station data analysis software and a capillary column model number Restek Rtx-CLPesticides, 30 m by 0.25 mm, 0.25 df column (maximum temperature 340 C, length 30.0 m diameter 250.0 μm, initial flow 2.0 ml/min, nominal initial pressure 22.39 psi, average velocity 43 cm/s) were used.

Lethal Effects of Treated Soil. Soil was collected as described above. Twenty grams of soil was placed in a 90-mm glass petri dish and moistened to 15% with water. One hundred workers (third instars or older), as determined by size, and 10 soldiers were placed on the soil. Treatments consisted of soils obtained from the four treated buildings (Ad. Cove, U.C., and C.A.).

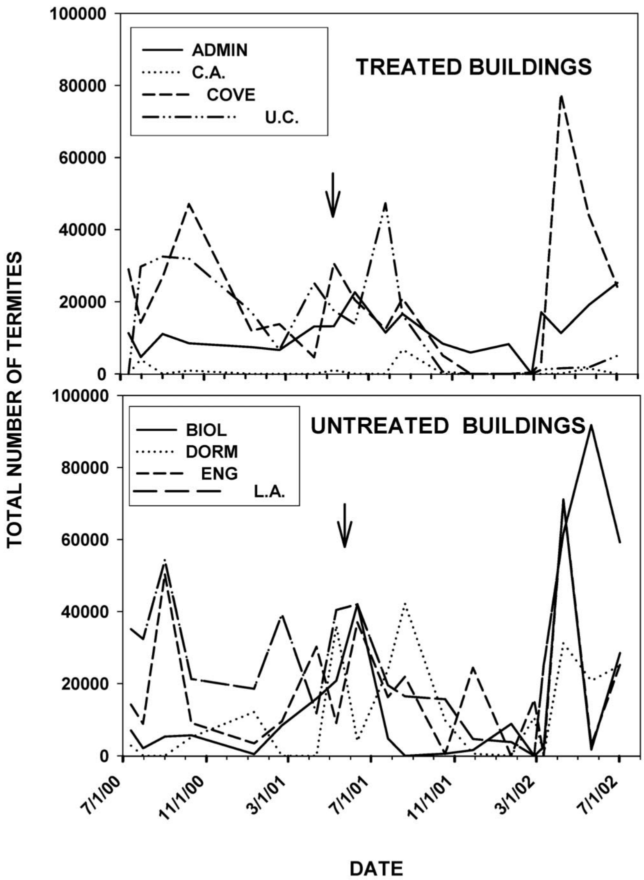

Fig. 2. Total number of *C. formosanus* workers trapped around treated and untreated buildings. Treated May 2001 (arrow).

Table 1. *C. formosanus* mean number of workers × 10^3 (grand mean ± SE) trapped at building sites

Date	No. collections (mo)	Treated				Untreated			
		Ad.	Cove	U.C.	C.A.	Biol.	Dorm	Eng.	L.A.
No. monitors		5	15	6	4	3	8	4	12
Pretreat 2000	7 (4.3)	1.4 ± 1.5	1.2 ± 1.1	1.8 ± 0.7	0.5 ± 0.4	2.6 ± 1.4	0.1 ± 0.4	4.1 ± 2.1	1.9 ± 1.2
Pretreat 2001	3 (3.5)	1.5 ± 1.6	0.7 ± 0.8	1.9 ± 2.3	0.0 ± 0.0	2.8 ± 1.5	0.0 ± 0.0	3.4 ± 0.6	1.9 ± 2.3
Posttreat 2001	6 (7)	2.6 ± 1.8	1.0 ± 1.3	2.5 ± 0.8	0.3 ± 0.5	3.7 ± 1.4	2.4 ± 2.0	4.6 ± 2.8	1.8 ± 1.2
Posttreat 2002	6 (6)	2.7 ± 2.5	1.7 ± 1.5	1.9 ± 2.1	0.1 ± 0.1	6.1 ± 5.3	1.4 ± 1.8	4.7 ± 4.3	3.2 ± 2.3
		$F = 0.123$	$F = 0.091$	$F = 0.052$	$F = 0.285$	$F = 0.285$	$F = 0.582$	$F = 0.023$	$F = 0.156$
		df = 3, 106	df = 3, 326	df = 3, 128	df = 3, 84	df = 3, 62	df = 3, 172	df = 3, 84	df = 3, 260
		$P = 0.947$	$P = 0.965$	$P = 0.984$	$P = 0.836$	$P = 0.836$	$P = 0.628$	$P = 0.995$	$P = 0.926$

An additional treatment of soil from the Cove, spiked with 50 ppm (wt:wt) imidacloprid was included. Spiked soil was prepared as described above. The acetone was allowed to evaporate from the substrate with periodic stirring over a period of 7 d. Controls (untreated substrates) consisted of potting soil and sand, respectively. It has been previously determined that acetone treatments of substrates have no discernible effect on termites after the acetone has evaporated. Each treatment was replicated four times with termites originating from four different colonies. Cumulative mortality (mean percentage and SD) was calculated for each treatment. Treatments from the same time were compared using ANOVA after transformation by the arcsine square root proportion mortality. Means were separated using Fisher's least-significant difference (LSD) multiple range test ($P = 0.05$) (SAS Institute 1990). Actual percentage of mortality is reported in the tables.

Termite Penetration into Treated Substrates. Bioassays were conducted in glass tubes (1.4 cm i.d. by 15 cm in height) with 5-cm segments of a centrally placed substrate contained on each end with 1 cm of 7% agar (Su and Scheffrahn 1990). Two wooden sticks and a strip of filter paper were placed into the 5-cm space at the bottom of the vertically placed tube. Fifty workers (third instars or older), as determined by size, and five soldiers were placed in the bottom space of four similarly prepared glass tubes (replicates). Filter paper was placed in the top void of the tube. Tubes were sealed at both ends with plastic caps and aluminum foil modified with pinholes for aeration. Substrates from the four treated buildings were tested along with control substrates of both sand and soil. Additionally, both sand and soil substrates containing 0, 1, 5, and 50 ppm imidacloprid were tested (wt:wt). Imidacloprid was added to the substrates in acetone, and the acetone was then allowed to evaporate with periodic stirring as described above. Substrate moisture was adjusted to 15%. Control substrates were moistened but not treated with insecticide. Substrates tested were sand (Standard Sand and Silica Company, Davenport, FL) and potting soil (Scotts Company, Marysville, OH). Organic matter in potting soil was ≈7% as determined by the Walkly-Black (Jackson 1958) wet digestion method (Louisiana State University Soil Testing Laboratory, Baton Rouge, LA) with 3.3, 66.7, and 30.0% sand, silt, and clay, respectively. Substrates were chosen because of their different affinities for absorption and adsorption of insecticides (Harris 1972). These substrates were near neutral (≈pH 6), as determined qualitatively with pH paper. Samples were held at 26.7 ± 1°C and ≈100% RH. Termite penetration (0–5 cm) was evaluated, and termite mortality was estimated daily for 7 d with the treated building substrates, and 13 d for the dose–response sand and potting soil substrates. Absolute termite mortality was determined at the end of the experiments, 7 or 13 d.

Cumulative penetration and mortality (mean percentage and SD) were calculated for each treatment. Treatments from the same time were compared using ANOVA after transformation by the arcsine square root proportion penetration or mortality. Means were separated using Fisher's LSD multiple range test ($P = 0.05$) (SAS Institute 1990). Actual percentage of penetration or mortality is reported in tables and figures.

Table 2. Mean ± SE (%) active traps at building sites

Date	No. collections (mo)	Treated				Untreated			
		Ad.	Cove	U.C	C.A.	Biol	Dorm	Eng.	L.A
No. monitors		5	15	6	4	3	8	4	12
Pretreat 2000	7 (4.3)	45.7 ± 72	41.0 ± 9.0	47.6 ± 11.2	14.3 ± 5.1	85.7 ± 6.7	3.5 ± 2.3b[a]	57.1 ± 10.5	54.2 ± 7.7
Pretreat 2001	3 (3.5)	53.3 ± 6.7	40.0 ± 7.7	61.1 ± 5.6	0.0 ± 0.0	99.9 ± 11.1	0.0 ± 0.0b	75.0 ± 14.4	66.7 ± 4.8
Posttreat 2001	6 (7)	53.3 ± 4.2	34.5 ± 8.3	61.1 ± 13.4	8.3 ± 5.3	61.1 ± 18.1	29.2 ± 5.3a	83.3 ± 8.3	52.8 ± 6.7
Posttreat 2002	6 (6)	50.0 ± 10.0	31.1 ± 8.7	30.6 ± 10.9	12.5 ± 5.6	55.6 ± 11.1	29.2 ± 7.7a	41.7 ± 14.0	48.6 ± 15.6
		$F = 0.230$	$F = 0.277$	$F = 1.408$	$F = 1.017$	$F = 2.217$	$F = 8.289$	$F = 2.554$	$F = 0.786$
		df = 3, 18	df = 3, 18	df = 3, 18	df = 3, 18	df = 3, 18	df = 3, 18	df = 3, 18	df = 3, 18
		$P = 0.875$	$P = 0.841$	$P = 0.273$	$P = 0.408$	$P = 0.121$	$P < 0.001$	$P = 0.088$	$P = 0.786$

Means followed by the same letter are not significantly different ($P < 0.05$; LSD).

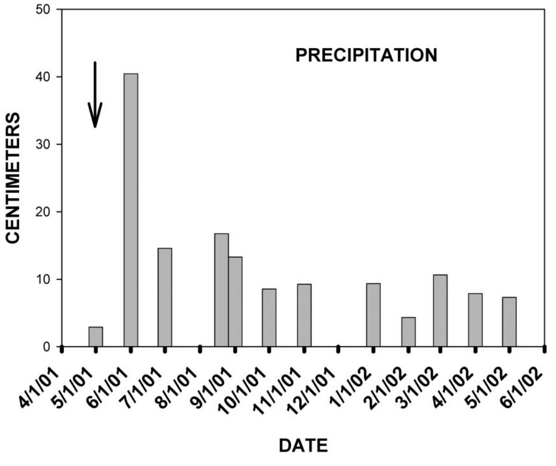

Fig. 3. Monthly precipitation posttreatment. Treated May 2001 (arrow).

Results and Discussion

Independent Monitors. None of the diagnostic behavioral changes and latent mortality attributed to imidacloprid intoxication were observed from *C. formosanus* collected from monitors and maintained in the laboratory (Boucias et al. 1996, Ramakrishnan et al. 2000, Thorne and Breisch 2001, Osbrink and Lax 2003). No obvious difference occurred in the total number of termites captured from independent monitors pre- or posttreatment in treated and untreated buildings (Fig. 2). No significant difference occurred in the grand mean number of termites ($F = 0.294$; df = 1, 55; $P = 0.590$) termites trapped posttreatment 2001 when comparing treated (1,325.8 ± 1,344.8) with untreated (2,590.5 ± 1,953.6) buildings. No significant difference occurred in the mean percentage of active traps ($F = 0.107$; df = 1, 46; $P = 0.746$) posttreatment 2001 when comparing treated (31.0 ± 24.7) with untreated (43.8 ± 30.1) buildings. No significant difference occurred in mean number of workers trapped around treated or untreated buildings when pretreatment counts are compared with posttreatment counts (Table 1). There were no significant reductions in mean percentage of active traps around treated and untreated buildings demonstrated before and after the treatments except in the case of the Dorm in which there was a significant increase in the percentage of active traps (Table 2). Thus, imidacloprid soil treatments did not measurably reduce *C. formosanus* populations adjacent to the treatments. These results are similar to the findings of Potter and Hillery (2002).

Soil Residue. After 1 yr, imidacloprid residues were not detectable (at 0.05 ppm) in soil collected from the Ad. and U.C. (Table 3). Imidacloprid was detected at low levels in the soil from the C.A. (0.08 ppm) and Cove (1.54). The analytical protocols are validated by the recovery of imidacloprid from Cove soil at 40 ppm when it was spiked with 50 ppm imidacloprid (Table 3). Wagner et al. (2005) reported loss of imidacloprid activity in Mississippi after 1 yr in the Forest Service ground board test at twice the rate applied in this study. Imidacloprid's 510 ppm water solubility is much higher than that of other termiticides (Nemeth-Konda et al. 2002). Possibly the ≈60 cm of rainfall in 2001

Table 3. Insecticide residues from soil around treated buildings

Bldg	Insecticide residue (ppm)						
	Date	Imidacloprid	Permethrin	Cypermethrin	Fipronil	Chlorpyrifos	Chlordane
Ad.	5–02	ND @ 0.05	ND@0.14	ND @ 0.14			21.30
U.C.	5–02	ND @ 0.05	ND@0.14	ND @ 0.14			319.00
C.A.	5–02	0.08	ND@0.14	13.2			0.08
Cove	5–02	ND @ 0.05	ND@0.14	ND @ 0.14			
2.19	10–02	1.54	0.76	ND @ 0.14	ND @ 0.03	ND @ 0.20	2.19
Cove (imidacloprid spiked, 50 ppm)	10–02	40.00	6.19	ND @ 0.14	ND @ 0.03	ND @ 0.20	26.21

ND, nondetectable.

Table 4. Mortality of *C. formosanus* in plate test by using substrate from treated buildings

Treatment substrate	% mortality (mean ± SD)[a]				
	Day 1	Day 2	Day 3	Day 4	Day 5
Ad.	0b	86.8 ± 7.7b	100.0 ± 0.0a	100.0 ± 0.0a	100.0 ± 0.0a
Cove	0b	0c	7.5 ± 2.9c	20.0 ± 7.1c	100.0 ± 0.0a
U.C.	0.8 ± 1.5b	100.0 ± 0.0a	100.0 ± 0.0a	100.0 ± 0.0a	100.0 ± 0.0a
C.A.	0b	3.8 ± 7.5c	51.3 ± 45.5b	56.3 ± 43.1b	100.0 ± 0.0a
Cove (imidacloprid-spiked, 50 ppm)	8.5 ± 1.9a	100.0 ± 0.0a	100.0 ± 0.0a	100.0 ± 0.0a	100.0 ± 0.0a
Soil	0bc	0c	0c	0d	0b
Sand	0bc	0c	0c	0d	0b
	$F = 38.2028.98$	$F = 314.57$	$F = 43.28$	$F = 44.96$	$F > 500.00$
	df = 6, 21	df = 6, 21	df = 6, 21	df = 6, 21	df = 6, 21
	$P < 0.0001$	$P < 0.0001$	$P < 0.0001$	$P < 0.0001$	$P < 0.0001$

Means within a column with the same letter are not significantly different ($P = 0.05l$; LSD).

[a] One hundred workers (third instar or older) and 10 soldiers per replicate were used, with four replicates.

Table 5. Cumulative percentage of mortality of *C. formosanus* in building substrate tube test

Treatment substrate	% mortality (mean ± SD)[a]						
	Day 1	Day 2	Day 3	Day 4	Day 5	Day 6	Day 7
Ad.	0c	0c	6.3 ± 2.5c	35.0 ± 4.1c	75.0 ± 0.0c	95.0 ± 5.8ab	100.0 ± 0.0a
Cove	0c	2.5 ± 5.0c	5.0 ± 10.0c	28.8 ± 14.4c	75.0 ± 12.3c	91.3 ± 6.3b	100.0 ± 0.0a
U.C.	22.5 ± 9.6a	87.5 ± 25.0a	100.0 ± 0.0a	100.0 ± 0.0a	100.0 ± 0.0a	100.0 ± 0.0a	100.0 ± 0.0a
C.A.	6.3 ± 4.8b	6.3 ± 4.8c	7.5 ± 5.0c	31.3 ± 2.5c	80.0 ± 4.1c	100.0 ± 0.0a	100.0 ± 0.0a
Cove (imidacloprid-spiked, 50 ppm)	30.0 ± 14.1a	52.5 ± 26.0b	63.8 ± 30.4b	83.8 ± 19.7b	100.0 ± 0.0a	100.0 ± 0.0a	100.0 ± 0.0a
Soil	0c	0c	0c	0d	1.3 ± 2.5c	1.3 ± 2.5c	1.3 ± 2.5b
Sand	0c	0c	0c	0d	1.3 ± 2.5c	1.3 ± 2.5c	1.3 ± 2.5b
	$F = 28.98$	$F = 29.11$	$F = 37.86$	$F = 64.02$	$F = 224.11$	$F = 224.11$	$F = 601.29$
	df = 6, 21	df = 6, 21	df = 6, 21	df = 6, 21	df = 6, 21	df = 6, 21	df = 6, 21
	$P < 0.0001$	$P < 0.0001$	$P < 0.0001$	$P < 0.0001$	$P < 0.0001$	$P < 0.0001$	$P < 0.0001$

Means within a column with the same letter are not significantly different ($P = 0.05$; LSD).
[a] One hundred workers (third instar or older) and 10 soldiers per replicate were used, with four replicates.

(Fig. 3) may have contributed to the loss of imidacloprid around the treated buildings. Although Baskaran et al. (1999) claims moisture has little effect on imidacloprid degradation, little information exists on the fate of imidacloprid in soil (Cox et al. 1997, 1998). It was shown by Junior et al. (2004) that the highest concentration of imidacloprid was found in drain water during the first drainage event after application. Julien et al. (1996) also found detectable residues of imidacloprid in runoff water from test plots. Furthermore, microbial degradation also can remove imidacloprid from soil (Rouchaud et al. 1994, Leib and Jarrett 2003). Variability in the permethrin and chlordane residues found in the repeated Cove samples is attributed to the heterogeneous nature of soil (Table 3). Chlordane residues were detected in all soils obtained from around the treated structures, with the highest being 319 ppm chlordane obtained from the U.C.

Lethal Effects of Treated Soil. Mortality of 100% occurred on all soils collected from around the treated buildings (Table 4). The most rapid mortality oc-

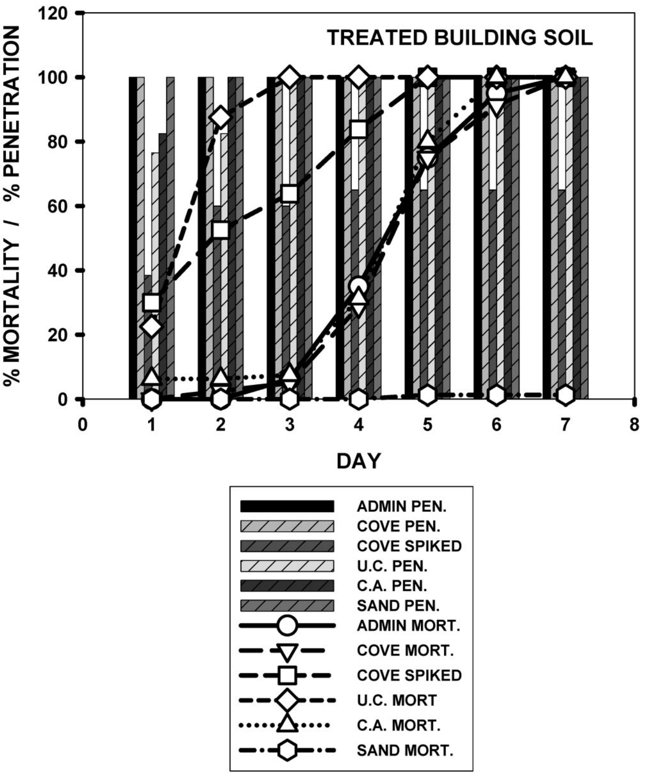

Fig. 4. Cumulative percentage of mortality and penetration of *C. formosanus* with soils obtained from perimeter of buildings 1 yr posttreatment with imidacloprid with sand as controls.

Table 6. Mean percentage of penetration by *C. formosanus* in building substrate tube test

Treatment substrate	% penetration (mean ± SD)[a]						
	Day 1	Day 2	Day 3	Day 4	Day 5	Day 6	Day 7
Ad.	100.0 ± 0.0a	100.0 ± 0.0a	100.0 ± 0.0a	100.0 ± 0.0a	100.0 ± 0.0a	100.0 ± 0.0a	100.0 ± 0.0a
Cove	100.0 ± 0.0a	100.0 ± 0.0a	100.0 ± 0.0a	100.0 ± 0.0a	100.0 ± 0.0a	100.0 ± 0.0a	100.0 ± 0.0a
U.C.	76.5 ± 21.8a	82.5 ± 20.6ab	100.0 ± 0.0a	100.0 ± 0.0a	100.0 ± 0.0a	100.0 ± 0.0a	100.0 ± 0.0a
C.A.	82.5 ± 23.6a	100.0 ± 0.0a	100.0 ± 0.0a	100.0 ± 0.0a	100.0 ± 0.0a	100.0 ± 0.0a	100.0 ± 0.0a
Cove (imidacloprid-spiked, 50 ppm)	38.5 ± 11.4b	60.0 ± 28.3b	60.0 ± 28.3b	65.0 ± 25.2b	65.0 ± 25.2b	65.0 ± 25.2b	65.0 ± 25.2b
Soil	100.0 ± 0.0a	100.0 ± 0.0a	100.0 ± 0.0a	100.0 ± 0.0a	100.0 ± 0.0a	100.0 ± 0.0a	100.0 ± 0.0a
Sand	100.0 ± 0.0a	100.0 ± 0.0a	100.0 ± 0.0a	100.0 ± 0.0a	100.0 ± 0.0a	100.0 ± 0.0a	100.0 ± 0.0a
	$F = 12.01$	$F = 5.29$	$F = 5.29$	$F = 8.46$	$F = 8.46$	$F = 8.46$	$F = 8.46$
	df = 6, 21	df = 6, 21	df = 6, 21	df = 6, 21	df = 6, 21	df = 6, 21	df = 6, 21
	$P < 0.0001$	$P = 0.0015$	$P = 0.0018$	$P < 0.0001$	$P < 0.0001$	$P < 0.0001$	$P < 0.0001$

Means within a column with the same letter are not significantly different ($P = 0.05$; LSD).
[a] Fifty workers (third instar or older) and five soldiers per replicate were used, with four replicates.

curred on soil from the U.C. (with >300 ppm chlordane [Table 3] and spiked soil [50 ppm imidacloprid] from the Cove, with 100% mortality by day 2.

Termite Penetration into Treated Substrates. As with the plate test, 100% mortality occurred in all tube tests containing soil from the perimeter of treated buildings with the most rapid mortality occurring in the U.C (Table 5; Fig. 4) with 100% mortality by day 3. Additionally, mortality in the imidacloprid-spiked Cove soil was delayed 3 d longer compared with forced exposure to the same treatment in the plate test. Thus, the presence of imidacloprid in a choice tube bioassay delayed the occurrence of mortality compared with the forced exposure of the plate test. There was 100% penetration through all substrates by 3 d with the exception of the imidacloprid-spiked Cove soil, in which only 65% penetration occurred (Table 6; Fig. 4). Thus, the termites were either avoiding contact with the imidacloprid-spiked Cove soil or the termites became intoxicated so rapidly that they stopped penetrating the soil. Imidacloprid intoxication has been reported to occur in termites within several hours (Boucias et al. 1996, Thorne and Breisch 2001).

The dose–response tube tests with imidacloprid by using two substrates showed 100% mortality only at the highest rate (50 ppm) after 13 d (Table 7; Fig. 5). Penetration was significantly inhibited at 5 and 50 ppm in sand and at 50 ppm in soil (Table 8; Fig. 5). Sand was only 40.5% penetrated and 5.5% penetrated at 5 and 50 ppm, respectively. Soil was penetrated 95.0 and 50.5% at 5 and 50 ppm imidacloprid, respectively (Table 8; Fig. 5). Sand retains more of the toxicant on the particle's surface than the other substrates (Osmun 1956, Harris 1972, Smith and Rust 1993, Osbrink and Lax 2003). Thus, although 50 ppm in the plate test caused 100% mortality in 2 d, in the tube choice test it took 13 d to cause 100% mortality. The presence of imidacloprid also inhibited the penetration of the substrates at 5 and 50 ppm. Thus, *C. formosanus* is detecting and/or being rapidly immobilized (within hours) by the presence of imidacloprid.

In this study, the inability of imidacloprid to have a detectable effect on termite populations within meters of the treatments, coupled with the inability of the termites to penetrate substrates treated even at low levels indicates that this compound does not fulfill the criteria to fit the model of being a slow-acting and nonrepellent termiticide (Potter and Hillery 2002, Thorne and Breisch 2001). The rapid rate of intoxication of termites by imidacloprid (Boucias et al. 1996, Thorne and Breisch 2001) makes it fast acting, even when mortality is delayed. It also must be noted that even if a chemistry fits the liquid-bait model of being nonrepellent and slow acting, the history of the substrate onto which it is applied must be taken into consideration. If a nonrepellent compound is placed

Table 7. Cumulative percentage of mortality of *C. formosanus* in imidacloprid-treated substrate tube test

Treatment substrate	Concn (ppm)	% mortality (mean ± SD)[a]						
		Day 1	Day 2	Day 3	Day 4	Day 6	Day 10	Day 13
Sand	0.0	0d	0d	0d	0d	0c	0c	0c
	1.0	3.5 ± 3.1cd	3.5 ± 3.1cd	6.3 ± 5.3cd	7.5 ± 3.7cd	13.8 ± 11.3bc	17.8 ± 11.9bc	41.5 ± 43.9bc
	5.0	7.8 ± 4.7bc	16.3 ± 11.3bc	17.5 ± 9.1bc	17.5 ± 9.1bc	30.5 ± 33.5abc	36.5 ± 42.5bc	57.8 ± 48.8ab
	50.0	27.3 ± 12.6a	46.5 ± 33.2a	54.8 ± 30.4a	54.8 ± 30.4a	62.3 ± 39.0a	87.8 ± 25.0a	100.0 ± 0.0a
Soil	0.0	0d	0d	0d	0d	0c	0c	0c
	1.0	0d	0d	0d	0d	0c	0c	0c
	5.0	0.3 ± 0.5d	1.5 ± 1.9d	2.3 ± 2.5d	3.3 ± 3.4d	4.5 ± 4.1c	21.3 ± 36.0bc	27.5 ± 48.5bc
	50.0	11.5 ± 4.8b	23.8 ± 5.3b	24.8 ± 6.2b	30.0 ± 8.2b	48.8 ± 27.8ab	62.5 ± 43.5ab	100.0 ± 0.0a
		$F = 22.87$	$F = 12.91$	$F = 18.02$	$F = 20.44$	$F = 8.75$	$F = 7.66$	$F = 10.48$
		df = 7, 24	df = 7, 24	df = 7, 24	df = 7, 24	df = 7, 24	df = 7, 24	df = 7, 24
		$P < 0.0001$	$P < 0.0001$	$P < 0.0001$	$P < 0.0001$	$P < 0.0001$	$P < 0.0001$	$P < 0.0001$

Means within a column with the same letter are not significantly different ($P = 0.05$; LSD).
[a] Fifty workers (third instar or older) and five soldiers per replicate were used, with four replicates.

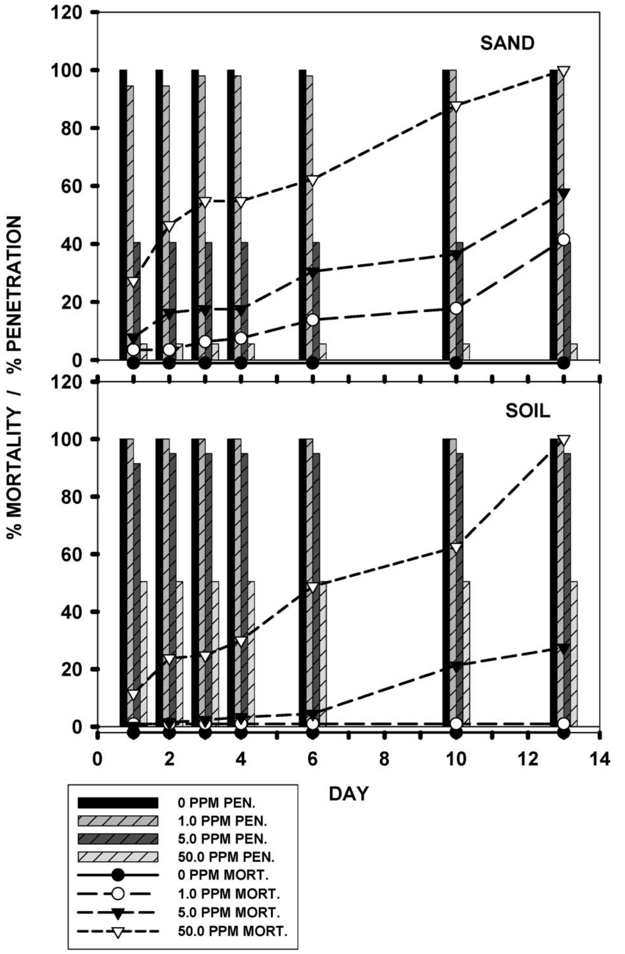

Fig. 5. Cumulative percentage of mortality and penetration of *C. formosanus* with two substrates treated with 0, 1.0, 5.0, and 50.0 ppm imidacloprid.

onto a substrate that contains repellent residues, the substrate will remain repellent. Another factor that has contributed to the success of some chitin synthesis inhibitors for termite population management and must be included in the liquid-bait model is the importance for the lethal time of an active ingredient to be dose independent (Su 2003; Su et al. 1982, 1995). The amount of toxicant absorbed by the termite is dependent on residence time spent in contact with the treated substrate, the amount of bait eaten, or the

Table 8. Mean percentage of penetration by *C. formosanus* of imidacloprid-treated substrate tube test

Treatment substrate	Concn (ppm)	% penetration (mean ± SD)[a]						
		Day 1	Day 2	Day 3	Day 4	Day 6	Day 10	Day 13
Sand	0.0	100.0 ± 0.0a	100.0 ± 0.0a	100.0 ± 0.0a	100.0 ± 0.0a	100.0 ± 0.0a	100.0 ± 0.0a	100.0 ± 0.0a
	1.0	94.5 ± 6.8a	94.5 ± 6.8a	98.0 ± 4.0a	98.0 ± 4.0a	98.0 ± 4.0a	100.0 ± 0.0a	100.0 ± 0.0a
	5.0	40.5 ± 22.3b	40.5 ± 22.3b	40.5 ± 22.3b	40.5 ± 22.3b	40.5 ± 22.3b	40.5 ± 22.3b	40.5 ± 22.3b
	50.0	5.5 ± 8.4c	5.5 ± 8.4c	5.5 ± 8.4c	5.5 ± 8.4c	5.5 ± 8.4c	5.5 ± 8.4c	5.5 ± 8.4c
Soil	0.0	100.0 ± 0.0a	100.0 ± 0.0a	100.0 ± 0.0a	100.0 ± 0.0a	100.0 ± 0.0a	100.0 ± 0.0a	100.0 ± 0.0a
	1.0	100.0 ± 0.0a	100.0 ± 0.0a	100.0 ± 0.0a	100.0 ± 0.0a	100.0 ± 0.0a	100.0 ± 0.0a	100.0 ± 0.0a
	5.0	91.5 ± 17.0a	95.0 ± 10.0a	95.0 ± 10.0a	95.0 ± 10.0a	95.0 ± 10.0a	95.0 ± 10.0a	95.0 ± 10.0a
	50.0	50.5 ± 9.2b	50.5 ± 9.2b	50.5 ± 9.2b	50.5 ± 9.2b	50.5 ± 9.2b	50.5 ± 9.2b	50.5 ± 9.2b
		$F = 37.98$	$F = 47.06$	$F = 53.54$	$F = 20.44$	$F = 53.54$	$F = 62.30$	$F = 62.30$
		df = 7, 24	df = 7, 24	df = 7, 24	df = 7, 24	df = 7, 24	df = 7, 24	df = 7, 24
		$P < 0.0001$	$P < 0.0001$	$P < 0.0001$	$P < 0.0001$	$P < 0.0001$	$P < 0.0001$	$P < 0.0001$

Means within a column with the same letter are not significantly different ($P = 0.05$; LSD).
[a] Fifty workers (third instar or older) and five soldiers per replicate were used, with four replicates.

amount of chemical transferred through the colony, and is beyond the control of the applicator of the treatment (Su 2003). Continued research into the most cost effective and environmentally friendly management strategies to suppress or eliminate termite populations should continue.

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